REMARKS

Currently pending in the application are claims 1-16, 18-25 and new claims 26-31. Claims 17 and 26 have been cancelled.

The Applicant's attorney of record points out that the Office Action was sent to the address of assignee Genencor International, Inc. It instead should have been sent to the attorney of record, the correspondent in the Declaration and Power of Attorney. It is requested that future correspondence be directed to the attorney of record, whose correspondence information is set forth at the end of the Amendment.

The Examiner did not receive the page containing the Abstract of the application and the Applicant has amended the specification to include an Abstract.

The claims have been objected to, with the Examiner requesting that the claim numbers not be separated from the text. The amendment incorporates the Examiner's request.

Claims 1-26 are rejected under section 112, second paragraph as being indefinite. The Applicant has adopted the Examiner's suggestions for claim changes, including adding the term "operably" before the term "linked" in claims 1 and 15, and changing "effect" expression to "control" expression. The Examiner also queries whether production levels of 0.01% TSP are considered to be commercial quantity. As discussed at page 24, lines 13-21 of the specification, levels of 0.01% are useful, with levels of 0.1% or more preferred. Claims 7-8 have been amended to clarify the language, indicating that the nucleotide sequence is a fungal or *Trametes versicolor* nucleotide sequence. Similar changes have been made to claims 20, 21 and 24. In claims 9 and 11 "producing" has been replaced with "encoding." At claims 11 and 24 the term "highly" has been added before the term "stringent." Please note that highly stringent conditions are discussed at length at pages 6-8 of the specification. In claims 16 and 17 "further" has been added before "comprising" and "a" substituted for "the" before the recitation of a globulin promoter in claims 12 and 19. Claim 22 has been amended to recite introducing a construct into a plant. Claims 25 has been rewritten to help avoid confusing terms and recites a method to produce laccase in commercial quantities by providing a biomass of plants, the plants having introduced therein laccase-encoding sequences expressing at levels of at about 0.01% or more.

Claims 13 and 14 have been rejected under section 101 as directed to non-statutory subject matter, the Examiner indicating the seed of claim 13 and the plant cells of claim 14 should include reference to the seed and plant being "transformed." However, the Applicant points out that both claims depend from claim 1 which recites a transformed plant, and thus are transformed seeds or cells.

Claims 1-26 are rejected under section 112, first paragraph, the Examiner indicating the claims are enabled for producing laccase at 0.1% total soluble protein in maize using SEQ ID NO: 1 under control of the maize globulin promoter and the barley amylase signal sequence, but not to other plants, at higher levels and using other laccase encoding genes. The Applicant respectfully traverses the rejection on several grounds.

The aim of the enablement requirements of section 112 of the patent laws is to assure that the Applicant has taught the invention through the disclosure of the specification such that one skilled in the art can understand and reproduce the invention. It is well accepted that it is not necessary to disclose each possible permutation of an invention. Here, the heart of the invention is the ability to successfully produce laccase in plants at higher levels than have ever been achieved. This was not possible before because the plants either expressed laccase at low levels (typically far below 0.01% total soluble protein), or the plants died. Indeed, that is also the experience of the inventors here. Accompanying this Amendment is the Declaration of Dr. Elizabeth Hood, a plant scientist with twenty years experience in the field of production of proteins in plants, and an inventor of the present invention. In her declaration, and the publication which is attached to the declaration, Hood et al., "Criteria for high-level expression of a fungal laccase gene in transgenic maize" *Plant Biotechnology Journal* (2003) 1:129-140, she states she has conducted a number of experiments in which laccase was expressed constitutively, or was not expressed outside the cell wall (see p. 140). Invariably, the expression levels were too low, or, when the expression levels reached higher amounts, the plants died. For whatever reason, the laccase enzyme appears to be toxic to the plant at high expression levels. Here, however, the inventors have achieved high expression levels by directing expression of the laccase to the seed of the plant, and preferably by expressing to the cell wall. In this manner the plant does not die and is capable of expressing high levels of laccase. This has not been possible before now.

Thus, which seed preferred promoter or which signal sequence is used is not of importance to the invention. The specification discusses many choices in promoter or signal sequences available to one skilled in the art. At page 8 beginning at line 14 of the specification the barley alpha amylase signal sequence is discussed, along with the KDEL sequence. The phaseolin promoter is another example of a seed-preferred promoter (p.10, line 35). There are dozens of such promoters and signal sequences available to the person skilled in this area. Which one is not the heart of this invention; that it is used is the key.

Nor is it critical which particular laccase-encoding sequence is employed. That the sequence encodes laccase is important, and many such sequences are known. In the specification among the examples provided are: the gene of laccase I cloned from *Aspergillus nidulans* as reported in Aramayo and Timberlake, *Nucleic Acids Res.* 18:3415 (1990); a gene from *Myceliophthora termophila* discussed by Berka et al, *Applied and Environmental Microbiology* p.3151-3157 (Aug. 1997).; a laccase gene from eucalyptus and pine for use in controlling lignin content in the plants described in PCT/NS97/00112; a laccase-encoding tobacco gene shown at WO 97/45549; a laccase-encoding gene corresponding to a *Rhizoctonia solani* gene set forth in 5,480,801; and a gene from a basidiomycete, *Polyporus pinsitus* discussed in U.S. patent 5,667,531. (See page 4, lines 8-21).

The Applicant points out to the Examiner that many variations in cell wall expressing sequences, seed-preferred expressing promoters and laccase genes are well known and available for selection to the scientist. Applicant has discussed such variations in the specification. Applicant did not produce in the specification each failed experiment where expression of laccase other than to the seed and not using cell-wall secreting sequences did not work. (This information is provided in the Declaration and attached publication of Dr. Hood). They have taught one skilled what to do.

Additional experiments following the methodology of the invention described in the specification have been carried out and have shown that expression levels in excess of 0.1% are achieved. In the Declaration of Dr. Hood, she states that experiments following such procedures have resulted in expression levels of 0.8% total soluble protein in T_1 seed. The results are set forth in the *Plant Biotechnology Journal* publication attached to

her Declaration. The ability to achieve these expression levels are also discussed in the second journal article attached to her Declaration, Bailey et al., "Improved recovery of active recombinant laccase from maize seed" *Applied Microbiology and Biotechnoloy,* (2003), in press, where even higher levels of expression were seen. The T_1 seeds having highest expression levels were selected (see page 13, line 25-30 of the specification), grown and backcrossed from Hi-II germplasm into another plant (the technique is described in the specification at page 15 beginning at line 22), and expression levels of at least about 10% total soluble protein are seen. Thus this invention does achieve high expression levels of laccase in plants for the first time.

The Examiner also says there is a lack of guidance for suitability of plants other than maize for high level expression of laccase in plants. However, there is as of the priority date of the application (1998) no reason to believe that plants other than corn cannot be transformed once transformation has been established. Dr. Hood's declaration includes her statement that it is a accepted that, absent evidence to the contrary, transformation of other plants is a matter of applying known methodology.

For these reasons it is respectfully submitted that the Applicant has enabled the invention and also has described the invention to convey to one skilled in the art that they had possession of the invention. Claims 1-26 are also rejected under section 112, first paragraph. The Examiner states the Applicant has not described a consensus sequence or structural element common to all laccase. However, the Applicant is not claiming a laccase-encoding gene as the invention. The Applicant claims use of any laccase-encoding gene and its high-level expression in plants. Any laccase gene, of which there are many, may be used. The sequence provided is merely one of many examples of sequences available to the scientist. Any variety of vectors may also be employed in the invention. By directing expressing to the seed of the plant, and preferably by directing expression to the cell wall, high levels of laccase expression may be achieved. Thus it is submitted the written description requirement is fulfilled, as is further evidenced by the experiments following the procedures of the specification which result in high levels of laccase expression in plants, as described in the publications and Declaration of Dr. Hood.

Claims 1-5, 11, 13-15 and 24-26 are rejected under section 102(a) as anticipated

by Bloksberg et al. The Examiner says that Bloksberg et al teaches tobacco plants transformed with DNA sequences encoding laccase, and notes that no expression levels are recited. The Examiner makes the assumption that the expression levels must be 0.01%, 0.1%, 1% and 10%, saying that since the LAC gene promoter and the globulin promoter are comparable in strength, such productions levels must be achieved. The Applicant respectfully traverses the rejection in that no expression levels are provided in the application, and any particular expression level cannot be presumed. Importantly, laccase in the Bloksberg application is expressed in leaf and stem. Bloksberg suggests that to increase lignin content, more copies of the gene are added, to decrease content, the antisense is introduced. However, as discussed *supra* and in Dr. Hood's declaration and the publications, adding more laccase genes will not result in higher expression.

In the experiments of the Bloksberg application cited by the Examiner, at page 18, line 25, in discussing the table on page 19, it is noted that the sense and antisense constructs of O-methyltransferase all resulted in significant decrease in levels of lignin (See also pp 20 and 21, Table 2; p.23, Table 3; p.24, Table 4 of Bloksberg). Table 4 shows decreased enzyme activity relative to control (empty vector transformed) plants. Both Bloksberg and the Faye application, cited for a section 102(b) rejection, do not show the invention where high levels of laccase expression, that is above 0.01% total soluble protein, and in particular above 0.1% TSP and up to 10% TSP are achieved. Nor does it show seed-preferred expression or cell-wall secretion. It is useful to understand that this application does not and cannot show the invention because they are trying to actually achieve a quite different goal. They are attempting to alter native lignin in plants by using certain constructs. They are down-regulating the gene in the pathway to reduce lignin. The control plants have the highest level of expression because the author's desire is to down-regulate, not obtain higher expression. Their goal is the quite different, they do not show seed-preferred, or cell wall secretion nor can they show high level expression of laccase in plants.

For these reasons the section 102(b) rejection of claims 1-5, 11, 13-15 and 24-26 is respectfully traversed. Faye et al teaches transformation of plants with sequences encoding laccase using *nos* and 35S promoter. Again, their goal is different, and they do not teach using seed-preferred promoters or signal sequences allowing the plant to express laccase at high levels.

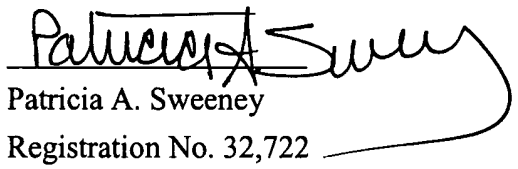
Claims 1-11, 13-18, and 20-26 are rejected under section 103(a) over Rodriguez in view of Ong et al. The Examiner says that Rodriguez teaches production of heterologous proteins in monocot plants and that plants, especially cereals including corn are excellent alternatives. The Examiner states Rodriquez does not teach laccase production in corn plants. The Examiner says that Ong teaches an isolated laccase-encoding DNA from *Trametes versicolor* and it would be obvious to combine the two. The Applicant agrees that showing transformation of a plant with a gene that expresses a particular protein in the plant shows that it can be introduced into other plants successfully. However, the Examiner has also pointed out that until introduction of the gene into a plant is achieved, results are unpredictable. High-level expression of laccase has not been achieved until now, but is now possible by keeping the plant alive through use of seed-preferred expression and cell wall targeting.

Claims 1-11, 13-15 and 20-26 are rejected under section 103(a) as unpatentable over Bloksberg in view of Ong for similar reasons, in that Bloksberg teaches transforming tobacco plants with a laccase gene but does not teach a gene from *Trametes versicolor*. The source of the gene, however, is not the key of the invention or claims. Bloksberg and Ong together cannot teach the contributions of this invention in expressing laccase in plants, not for modification of lignin biosynthesis to down-regulate, but to obtain higher level of expressions than can be achieved using the procedures of Bloksberg or Ong.

The Applicant respectfully requests that in the event there are further issues relating to allowance of the claims, that a telephonic interview be granted.

For the foregoing reasons, reconsideration and allowance of the claims is requested.

Respectfully submitted,



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